281

Signaling networks via the epithelio-mesenchymal interactions during early development of mouse tongue

Yuji Taya, Takako Hirano, Kaori Sato, Takaaki Aoba
Dept. of Pathol., Nippon Dental Univ., Tokyo, Japan

We have found that the muscle progenitor cells contacted with the epithelium beneath the median sulcus of lateral lingual swellings in early tongue development. We focused on the molecular regulation of mandibular process—fusion and subsequent tongue morphogenesis via epithelio-mesenchymal interactions. Samples for gene expression were obtained from the tongue primordium, or its mesenchymal cell populations that were collected using laser microdissection, in ICR mouse embryos at E9.5–12.5. mRNA expression profiles were analyzed in combination of microarray and real-time PCR. A bundle of genes such as ET1, Bmp4, Msx1, Dlx6, dHand, and Follistatin disclosed distinct spatiotemporal expression patterns. ET1 expression increased transiently in the epithelium of the medial region of the mandibular arch at E9.5, while ET1-related genes were expressed in the mesenchyme, supporting the activation of signaling pathways ('ET1'–'Dlx6'–'dHand'–'Msx1') in epithelio-mesenchymal interactions during the mandibular fusion. At the stage of the formation of lateral lingual swellings, BMP4 expression was increased markedly, supporting the function of Bmp4 for the proliferation of the mesenchymal cells in lateral lingual swellings. In the medial region of the tongue primordium at E11.5–12.5, expression of myogenic transcription factors MyoD and Myf5 appeared to be tightly regulated by counter balancing between Bmp4 and its antagonists Noggin and Follistatin. The overall results indicate that epithelium-derived signals such as ET1 and Bmp4 play pivotal roles in the medial fusion of mandibular arch and early development of the tongue primordium.

doi:10.1016/j.ydbio.2006.04.309

282

Endoderm patterning in embryos and in human ES cell cultures

James M. Wells, Billie A. Moore-Scott, Suh-Chin Jackie Lin, Robert Opoka
Cincinnati Children's Hospital Research Foundation, Cincinnati, OH, USA

Between the gastrulation and early somite stages, the endoderm germ layer is transformed from an undetermined sheet of cells to a primitive tube that is patterned along the A–P axis. Our studies show that endoderm patterning is highly conserved across vertebrate species and FGF signaling plays a central role in establishing these gut tube domain in vivo. The expression pattern of one ligand, FGF4, as well as functional studies, suggests that FGF signaling patterns the gut tube by promoting posterior and repressing anterior endoderm cell fate. We show that FGF4 concentration can directly regulate A–P endoderm cell fate and may also act to control the path of endoderm cell migration along the A–P axis, suggesting that both mechanisms act to pattern the developing gut tube. We have identified one endoderm-specific, FGF4-responsive gene that encodes FGF binding protein 1 (FGFbp1). Our studies have determined that FGFbp1 is secreted, binds to FGF4, and increases its solubility by preventing it from becoming tethered to the extracellular matrix (ECM) resulting in enhanced FGF4 bioavailability and bioactivity. These data support our hypothesis that FGFbp1 acts as an endoderm-specific FGF4 agonist during endoderm and gut tube patterning. We are using developmental paradigms to direct the differentiation of human embryonic stem cells into endoderm like cells, as well as patterning these cells into specific A–P lineages.

doi:10.1016/j.ydbio.2006.04.310

283

HBEGF prevents trophoblast apoptosis due to reoxygenation injury during placentation

D. Randall Armant [1], Brian A. Kilburn [1], Po J. Chiang [1], Anelia Petkova [1], Roberto Romero [2], Richard E. Leach [3]
[1] *Department of OB/GYN, Wayne State University, Detroit, MI, USA*
[2] *Perinatology Research Branch, NICHHD, NIH, DHHS, Bethesda, MD, USA*
[3] *Department of OB/GYN, University of Illinois, Chicago, IL, USA*

Human cytotrophoblast cells (CTC) survive well at reduced O_2 levels due to upregulation of heparin-binding EGF-like growth factor (HBEGF), a member of the EGF family that is deficient in the pregnancy disorder preeclampsia. It has been suggested that aberrant placentation resulting in uterine vasospasms causes reoxygenation injury of CTC, which undergo elevated apoptosis in preeclampsia. We have examined whether HBEGF signaling influences CTC survival in an in vitro hypoxia/reoxygenation (H/R) model using a human first trimester CTC line (HTR-8/SVneo) exposed to 2% O_2 for 2 h and then reoxygenated (20% O_2) for 6 h. H/R significantly increased cell death determined by TUNEL. Apoptosis was established by preventing cell death with caspase inhibitors and detecting annexin V binding to CTC. However, necrotic release of LDH from CTC was not observed. A specific ELISA showed that HBEGF, upregulated by 2% O_2, was downregulated within 1h of reoxygenation. CTC apoptosis was ameliorated by adding recombinant HBEGF or other EGF family members during H/R. HBEGF rescue was prevented by an HBEGF-specific antagonist or blocking antibodies against its receptors, HER1 and HER4. We conclude that the EGF signaling system provides an important survival mechanism for CTC and that downregulation of HBEGF that occurs in preeclampsia could contribute to the observed apoptosis of CTC. Supported by NIH grants HD98004 and HD37500 and the intramural research program of NICHHD.

doi:10.1016/j.ydbio.2006.04.311